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Comparing the Tadpoles of *Hyla geographica* and *Hyla semilineata*

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ABSTRACT.—External morphology, internal oral anatomy, and chondrocranial anatomy were examined for tadpoles of *Hyla geographica* from the Amazon rainforest, Brazil, and *Hyla semilineata* from the Atlantic rainforest, Brazil. Here, we provide morphological larval data to help diagnose these closely related species. Scanning electron microscopy analysis of buccal morphology showed the most distinctive features between these species: the distance between the lingual papillae in the buccal floor of *H. geographica* is three times greater than that distance in *H. semilineata*, and the relative size of the lingual papillae in *H. geographica* is less than half their size in *H. semilineata*. Although the chondrocranium of both species is identical, and the external morphology of the larvae of both taxa is very similar, they differ greatly in size at most developmental stages. A multivariate analysis of covariance, corrected for stage and size, also showed a significant difference between morphometric measurements of the two species. These differences support the existence of two separate taxa.

The identity of *Hyla geographica* and *H. semilineata* has not been satisfactorily resolved. *Hyla geographica* was described in 1824 by Spix, who also described a variety identified as *H. geographica* var. *sive semilineata*. These taxa were synonymized by Bokermann (1966). Subsequently, Lutz (1973) stated that there are “no clear-cut differences” between the two taxa, and Duellman (1973) also placed both taxa under the name *H. geographica*. More recently, Silveira and Caramaschi (1989) compared adult morphology and argued for the specific recognition of *H. semilineata* for the taxa inhabiting the coastal region of Brazil, from the State of Alagoas to the State of Santa Catarina. These authors differentiated *H. semilineata* from *H. geographica* by its narrower head, more rounded snout, smaller tympanum, larger nuptial pad, and color pattern.

General descriptions of tadpoles and their oral discs have been used as taxonomic characters sufficient for species differentiation (Bokermann, 1963; Altig, 1970). However, the descriptions of the larvae of *Hyla geographica* and *Hyla semilineata* in the literature are very similar, suggesting that a more detailed study of the oral disc and of other morphological characters may be important for distinguishing these species. The tadpoles of *H. geographica* and *H. semilineata* are classified as Orton type IV larvae (Orton, 1953), because they possess a horny beak furnished with denticles and a single, sinister spiracle. These tadpoles, like most hylids, are adapted to lentic environments. Characteristically, they possess an oval-shaped body, with a

tail about twice the size of the body, and an antero-ventral mouth. (Orton, 1953; Bokermann, 1963; Kenny, 1969; Duellman and Lescure, 1973; Duellman and Trueb, 1994). Bokermann (1963) described the tadpoles from the Atlantic rainforest (Piassaguera, State of São Paulo, Brasil) as being blue/black with gray limbs and having a dental formula of 2(2)/4. Kenny (1969) described the tadpoles from Trinidad in a similar manner as did Duellman and Lescure (1973) for the species from French Guiana (identified incorrectly as *Osteocephalus taurinus*; see Caldwell, 1989), although the latter observed a 3(1)/5 dental formula at Stage 37.

We examined larvae of *Hyla geographica* and *Hyla semilineata* from allopatric populations and here we describe their morphometric differences, external and internal oral structures, and chondrocranial anatomy, presenting features that help to distinguish the two species.

MATERIALS AND METHODS

Tadpoles of *Hyla geographica* were collected at the Reserva Florestal Adolfo Ducke, Manaus, State of Amazonas, Brasil in January, 1996. Tadpoles of *Hyla semilineata* were collected at the following localities: Poço das Antas, Mongaguá, State of São Paulo, Brasil in April, 1994 and October 1995; Picinguaba Beach, Ubatuba, State of São Paulo, Brasil in January 1992; Massaguaçu Beach, Caraguatatuba, State of São Paulo, Brasil, in September, 1993; and Recanto Silvestre, Blumenau, State of Santa Catarina, Brasil in January, 1996. Specimens are deposited in the CFBH collection of the Departamento de Zoologia, Instituto de Biociências, UNESP, Rio Claro, São Paulo, Brazil.

Tadpoles were preserved in 10% buffered formaldehyde solution. Most of the tadpoles

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were preserved shortly after collection, but some were reared in the laboratory to verify identification. Tadpoles were staged according to Gosner's table of normal development (Gosner, 1960). Measurements were taken with calipers to the nearest 0.1 mm. Morphometric measurements of 20 specimens of each species from Stage 31 to 39 follow Lavilla and Scrocchi (1986). Morphometric measurements were: total length (TL), body length, from snout to vent (BL), tail length, from vent to tip of tail (TaL), maximum body width (BW), body width at eye level (BWE), body width at nostril level (BWN), maximum body height (BH), maximum tail height (TaH), tail muscle height (TMH), rostrum-spiracular distance (RSD), frontonasal distance (FN), naso-ocular distance (NO), extraorbital distance (EO), interorbital distance (IO), extranasal distance (EN), internarial distance (IN), eye diameter (E), nostril diameter (N), and rostral gap size (RG).

Morphometric variation between the 40 larval specimens of *Hyla geographica* and *H. semilineata* was assessed by a principal component analysis (PCA) of the variance-covariance matrix of log-transformed values (Neff and Marcus, 1980). A PCA was also produced after Burnaby (1966) correction for size. To test for significance of shape differences between the two species, a multivariate analysis of covariance (MANCOVA) was undertaken controlling size and developmental Stage. The STATISTICA for Windows (StatSoft, Inc., 1996) computer program was used for the statistical analysis.

The external structure of the oral disc was examined in tadpoles between Stage 25 to 42. Seven specimens of each taxa were dissected for observation of the internal oral anatomy under scanning electron microscopy (SEM) and prepared as follows: The dissections were submitted to ultrasonic cleaning for 20 min. They were then fixed in 4% glutaraldehyde for 2 h at room temperature, followed by three 15 min washes with 0.1 M phosphate buffer and post fixed in 1% osmium tetroxide for 2 h room temperature. Three 15-min washes with 0.1 M phosphate buffer followed. Specimens were next dehydrated using 15 min changes of an ethanol series (35%, 50%, 70%, 80%, 95%, 3 × 100%). Specimens were critical point dried in CO₂, mounted on aluminum stubs and sputter coated with gold/palladium to 30 nm thickness, using a Hummer VII sputtering system. Internal oral anatomy was examined under a Hitachi S-2300 scanning electron microscope at 15kV and photographed using Polaroid 55 positive/negative film. Terminology of internal surface features follows Wassersug (1976).

Tadpoles were cleared and double-stained with Alcian Blue and Alizarin Red following

Dingerkus and Uhler (1977). Chondrocranium was observed under a stereomicroscope (Leica-Wild M3C) and drawings were made with the aid of a camera lucida. The description of the chondrocranium was based on larvae at Stage 30. Terminology follows de Sá (1988) and Haas (1995).

RESULTS

Descriptions apply to both species, unless otherwise specified.

External Morphometrics.—The first principal component (PC1) accounted for 83.3% of the variation in the sample. It was interpreted as a size factor because all the characters had a positive sign (indicating that the variables have the same direction of variation) and high correlation coefficients (Table 1, Fig. 1). The second principal component (PC2) accounted for only 5.8% of the variation between the specimens and had very low correlation coefficients. It was interpreted as a shape factor because the coefficients had both positive and negative signs (indicating that the variables have different directions of variation) (Table 1, Fig. 1). The second principal component analysis, carried out after Burnaby correction for size, produced very low correlation coefficients (Table 1, Fig. 2). The first principal component after Burnaby correction (PC1B) accounted for 34.6% of the variation in the sample and the second principal component (PC2B), for 28.9%. Both principal components had positive and negative signs and were interpreted as shape factors.

The chief interspecific difference between *H. geographica* and *H. semilineata* can be attributed to size because most of the scores of PC1 for *H. semilineata* have higher values than those for *H. geographica* (Fig. 1). This difference in size between the two species was constantly distributed along the developmental stages examined, the tadpoles of *H. semilineata* being about 1.5 times larger than those of *H. geographica* at most developmental stages.

There was a large overlap of values for the scores of PC2 and a considerable overlap in the scores of the principal component analysis after Burnaby correction (Fig. 1, 2). However, a difference related to shape can be seen in the scatterplot of PC2B (Fig. 2) where the scores for *H. semilineata* are generally higher than for *H. geographica*. Although the tadpoles of *H. semilineata* are relatively longer in total length and body length than *H. geographica*, their tail is shorter (positive TL and BL coefficients and negative TaL coefficient). *Hyla semilineata* is relatively wider and taller than *H. geographica* (positive BW, BWE, BWN, BH, FH, and TMH coefficients). The spiracle is located closer to the snout in *H. semilineata* (negative RSD coefficient), as

TABLE 1. Coefficients of the principal component analysis for 19 measurements taken for *Hyla geographica* and *Hyla semilineata* without correction for size (PC1 and PC2) and after Burnaby correction for size (PC1B and PC2B). See Figure 1 and 2.

Character	PC1	PC2	r (PC1)	r (PC2)	PC1B	PC2B	r (PC1B)	r (PC2B)
TL	0.1111	0.0082	0.98	0.07	0.0082	0.0044	0.07	0.04
BL	0.1097	0.0090	0.97	0.08	0.0090	0.0165	0.08	0.15
TaL	0.1125	0.0071	0.95	0.06	0.0071	-0.0043	0.06	-0.04
BW	0.1164	0.0127	0.98	0.11	0.0127	0.0098	0.11	0.08
BWE	0.1034	0.0038	0.98	0.04	0.0038	0.0106	0.04	0.10
BWN	0.1043	-0.0105	0.94	-0.10	-0.0105	0.0217	-0.10	0.20
BH	0.1340	0.0134	0.95	0.10	0.0134	0.0281	0.10	0.20
FH	0.1457	0.0236	0.96	0.16	0.0236	0.0258	0.16	0.17
TMH	0.1256	0.0199	0.92	0.15	0.0199	0.0132	0.15	0.10
RSD	0.0927	0.0161	0.95	0.17	0.0161	-0.0022	0.17	-0.02
FN	0.0932	-0.0123	0.94	-0.12	-0.0123	-0.0128	-0.12	-0.13
NO	0.0901	-0.0221	0.85	-0.21	-0.0221	-0.0250	-0.21	-0.23
EO	0.0840	0.0123	0.96	0.14	0.0123	-0.0091	0.14	-0.10
IO	0.1270	0.0140	0.96	0.11	0.0140	0.0075	0.11	0.06
EN	0.0614	0.0058	0.95	0.09	0.0058	-0.0094	0.09	-0.14
IN	0.0714	-0.0044	0.93	-0.06	-0.0044	0.0005	-0.06	0.01
E	0.0585	0.0465	0.51	0.41	0.0465	-0.0840	0.41	-0.73
N	0.0456	0.0275	0.59	0.35	0.0275	-0.0336	0.35	-0.43
RG	0.1695	-0.0967	0.85	-0.49	-0.0967	-0.0298	-0.49	-0.15

are the nares and eyes (negative FN and NO coefficients). The nares and eyes are relatively smaller in *H. semilineata* and positioned further apart (positive IN, negative EN and N coefficients; positive IO, negative EO and E coefficients). The rostral gap is relatively narrower in *H. semilineata* (negative RG coefficient).

The MANCOVA performed, controlling size (PC1) and developmental Stage, showed significant results (Wilks' Lambda criterion = 0.182; $P = 0.002$).

Structure of Oral Disc.—The mouth is ventral and two rows of marginal papillae border the oral disc, except for a rostral gap. Papillae are larger on the posterior margin and are spaced irregularly. There is individual variation in the size and spacing among these lower papillae. A lateral fold appears at Stage 26 and is clearly visible from Stage 29 until Stage 43, folding over the oral disc. The rostral gap is small and varies in size according to the Stage. The horny beaks are small and serrated; the upper beak is broad-

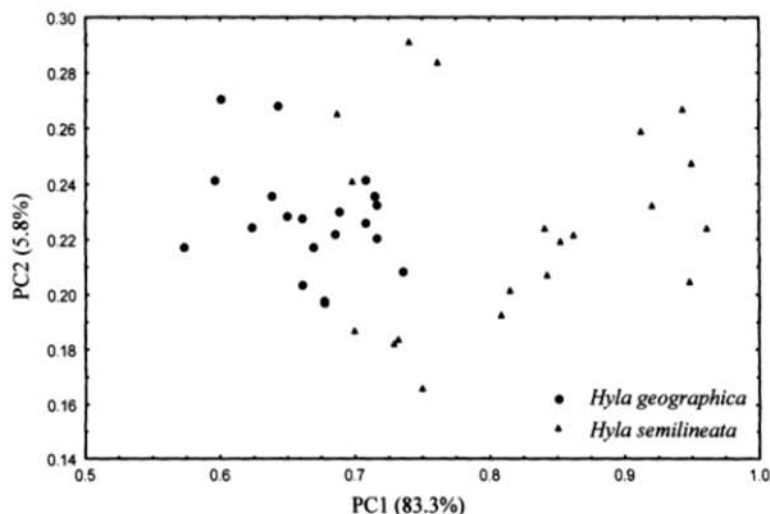


FIG. 1. Scatterplot of the sample of *Hyla geographica* and *H. semilineata* tadpoles in the space of the first two principal components. The numbers in parentheses are the percentage of contribution of each component to the total variance in the sample.

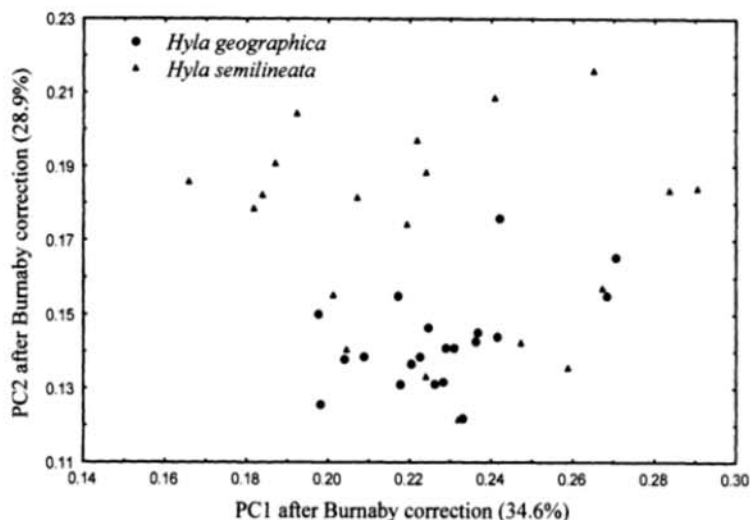


FIG. 2. Scatterplot of the sample of *Hyla geographica* and *H. semilineata* tadpoles after Burnaby correction. The numbers in parentheses are the percentage of contribution of each component to the total variance in the sample.

ly rounded whereas the lower beak is triangular. At Stage 41, the beaks are larger and the rows of papillae no longer fold over the oral disc. The second row of upper denticles is interrupted medially. The number of denticle rows varies depending on the individual. At Stage 25 the dental formula tends to be 2(2)/3 and may increase to 2(2)/4 and 2(2)/5 by Stage 28. Between Stages 27 and 30, one extra, medially interrupted upper row of denticles, could frequently be observed between the papillae under the lateral folds of the anterior margin. After Stage 31, the number of rows of the inferior denticles reduces to three or four. At Stage 41, only vestiges of the denticle rows can be seen. By Stage 42/43 the rows of papillae have reduced in size and denticles have disappeared.

Internal Oral Anatomy.—The buccal floor (Fig. 3) is about as wide as long. In the prelingual area, infrarostral papillae with postulated margins were observed, three pairs in *Hyla geographica* (Fig. 3a) and four pairs in *H. semilineata*. Two major, infralabial, palplike papillae extend medially from the lateral walls of the mouth. These are more than twice the size of the minor and more medial pair that project dorsally from the floor of the mouth. All have postulated margins. A pair of simple and slender lingual papillae are found close to the infralabial papillae. In *H. geographica*, the stubby lingual papillae (Fig. 3a) are less than half the length and are positioned further apart than in *H. semilineata*, where they are attenuate (Fig. 3b). The mean distance between the two lingual papillae in *H. semilineata* (0.04 mm; SD = 0.01; N = 3) is about 1/3 of that distance in *H. geographica* (0.13 mm;

SD = 0.02; N = 3). The buccal floor arena (BFA) is limited laterally and posteriorly by about 10 to 15 BFA papillae on each side, forming a semicircle. The BFA papillae are attenuate, usually simple with some bi- or trifurcate. Scattered postulations are found among the BFA papillae and also in the center of the BFA. About 10 pre-pocket papillae are dispersed anterior to the buccal pockets. Buccal pockets are about twice longer than wide and are located at a 30° angle from the transverse plain. There is a free, long, semi-circular velar surface, equal to the widest part of the buccal floor, with small peaks over the filter cavities and a distinct median notch. Dense secretory pits are found along the margin of the velar surface. The glottis is large, located below the velum (Fig. 3b).

In the roof of the mouth (Fig. 4), the prenarial area has a large horizontal papilla surrounded by scattered postulations, the number and size of which varied among the specimens examined. The nares are located 1/3 posteriorly in the roof, oriented antero-medially, with large postulations on the anterior wall and minor ones on the medial wall. The posterior wall is smooth-edged. The postnarial area has a pair of thick, attenuate, conical papillae, that bend medially. About six to eight smaller papillae and several postulations surround them. There is a pair of lateral ridge papillae with postulated anterior margins. The median ridge is triangular, its length equal to width, with a rugose edge. It is located about half way along the roof rostro-caudally. The buccal roof arena (BRA) is large and oval with numerous postulations. In general, two to three simple lateral roof papillae

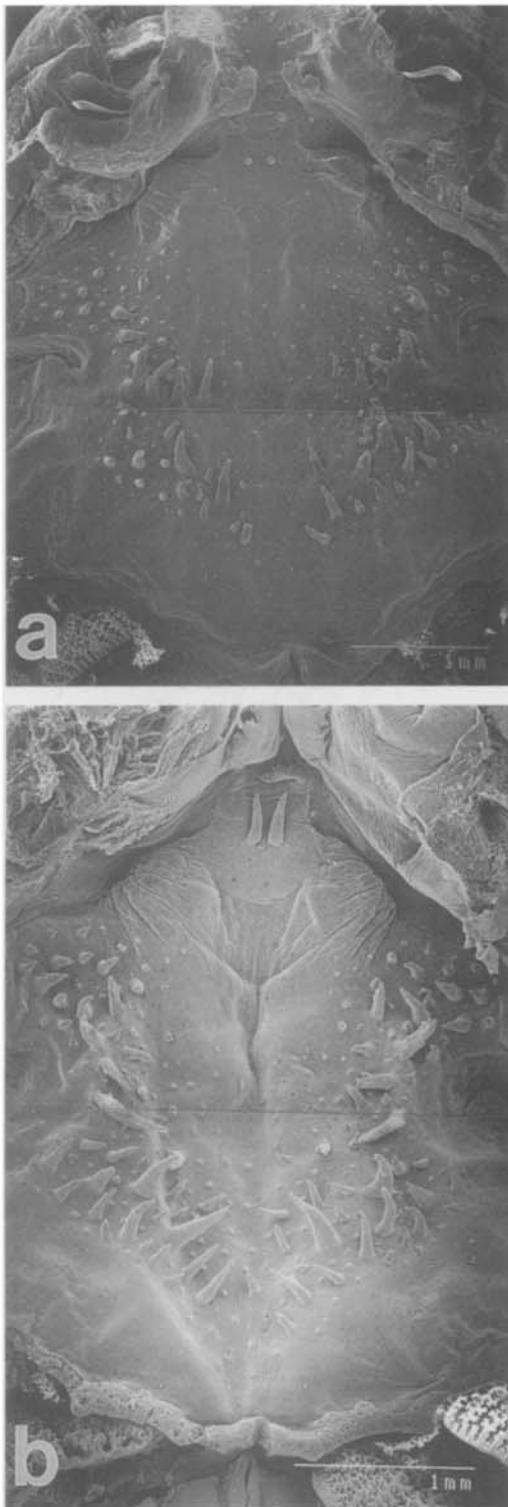


FIG. 3. Buccal floor of (A) *Hyla geographica*, Stage 38, TL = 42.43, and (B) *Hyla semilineata*, Stage 37, TL = 43.71. SEM micrograph at 15kV. Notice that the lin-

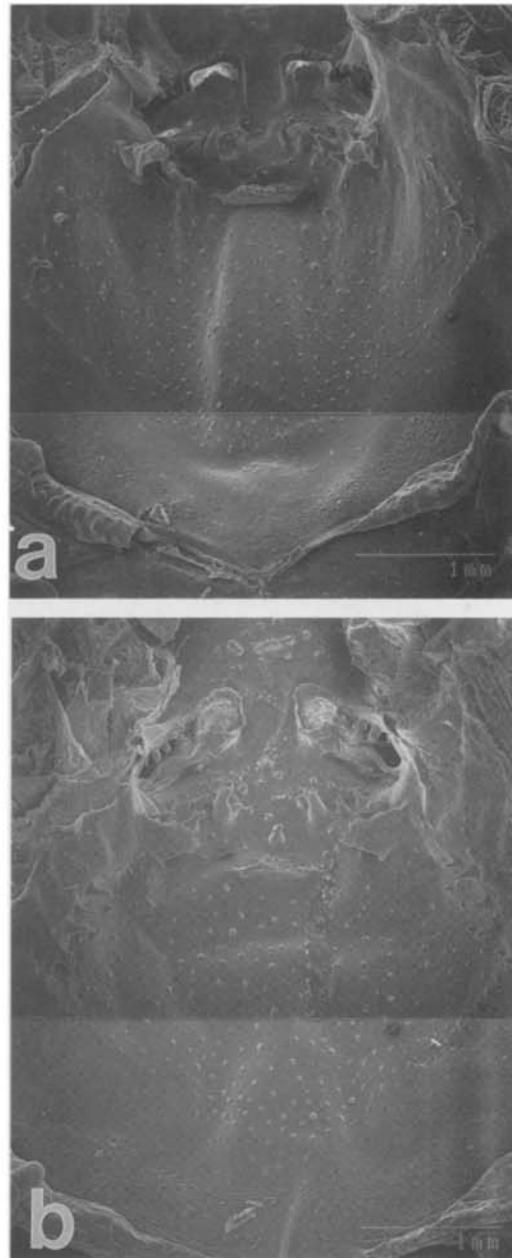


FIG. 4. Buccal roof of (A) *Hyla geographica*, Stage 38, TL = 42.43, and (B) *Hyla semilineata*, Stage 38, TL = 48.21. SEM micrograph at 15kV. Notice that there are two lateral ridge papillae in *H. geographica* and four in *H. semilineata*.

←

gual papillae are shorter in *H. geographica* and spaced further apart than in *H. semilineata*.

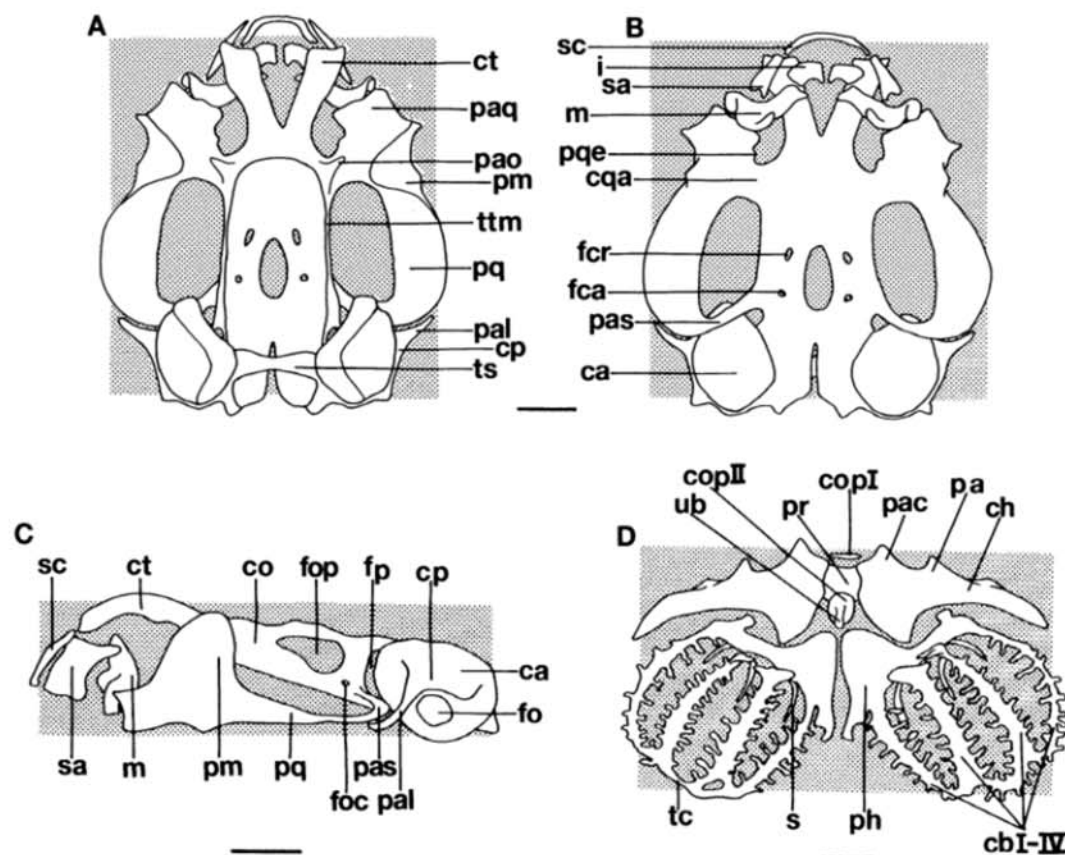


FIG. 5. Chondrocranium of *Hyla geographica* (Stage 30; CFBH# 2810). (A) Dorsal view, (B) ventral view, (C) lateral view, and (D) ventral view of hyobranchial apparatus. Abbreviations: ca = capsula auditiva, cb = ceratobranchial, ch = ceratohyal, co = cartilago orbitalis, copI = copula I, copII = copula II, cp = crista parotica, cqa = commissura quadrato cranialis anterior, ct = cornua trabeculae, fca = foramen caroticum primum, fcr = foramen craniopalatinum, fo = fenestra ovalis, foc = foramen oculomotorium, fop = foramen opticum, fp = foramen prooticum, i = cartilago infraorbitalis, m = cartilago Meckeli, pa = processus anterolateralis hyales, pac = processus anterior hyales, pal = processus anterolateralis, pao = processus antorbitalis, paq = pars articularis quadrati, pas = processus ascendens, ph = planum hypobranchiale, pm = processus muscularis quadrati, pq = palatoquadrate, ppe = processus quadrato ethmoidalis, pr = pars reuniens, s = spiculum, sa = pars alaris, sc = pars corporis, tc = commissura terminalis, ts = tectum synoticum, ttm = taenia tecti marginalis, ub = processus urobranchialis. Bar = 1 mm.

line the wall of the BRA in *H. geographica* (Fig. 4a), and three to four in *H. semilineata* (Fig. 4b). There is a large conspicuous glandular zone that extends onto the dorsal velum which is interrupted medially.

Chondrocranium.—At Stage 30, the chondrocranium is completely cartilaginous in *Hyla semilineata*, whereas in *H. geographica* the frontoparietal has begun to ossify.

The cartilago suprarostalis provides support for the upper horny beak, which functions as the upper jaw of the tadpoles. The slender corpora of the suprarostalis are attached ventromedially and lie in the anterior-most part of the chondrocranium, forming a flattened semi-circle ventral to the anterior ends of the cornua tra-

becularum with which they articulate posteriorly (Fig. 5B, 6). Slightly posterior, ventral to each corpora, and articulating proximally are the relatively broad and lateral alae of the suprarostalis. The alae are located ventrolaterally to the anterior halves of the cornua trabecularum (Fig. 5B, C). By Stage 43, the suprarostalis and the distal ends of the cornua trabecularum have disappeared in *Hyla geographica*, whereas in *H. semilineata* these structures disappear slightly later, at Stage 44.

The cornua trabecularum are about one quarter of the total chondrocranial length. They project anteriorly from the planum ethmoidale, diverging from one another in the shape of a "V", to form two rectangular bars that become slight-

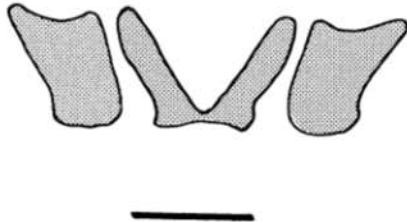


FIG. 6. Frontal view of the cartilago suprarostalis of *Hyla geographica* (Stage 29; CFBH# 2809). Bar = 1 mm.

ly wider in their distal half (Fig. 5A). They curve ventrally to articulate with the suprarostalis.

The lower jaw of the tadpole consists of the medial and paired cartilages infrarostrales, which support the lower horny beak, and the lateral paired cartilages Meckeli (Fig. 5B, C). The cartilages Meckeli are short and curved, placed ventral to the cornua trabecularum and posterior to the lateral alae of the suprarostalis. They articulate with the infrarostralis anteroventrally. The convex posterior margin of the cartilago Meckeli articulates broadly with the articular portion of the palatoquadrate so that the rounded tip of the lateral processus retroarticularis lies anteriorly to the pars articularis quadrati (Fig. 5A, B, C). By Stage 43, the posterolateral end of the infrarostralis is fused with the anteromedial end of the cartilago Meckeli which has begun to lengthen.

Posteriorly, the planum ethmoidale is continuous with the cranial floor. Dorsally, the posterior edge of the planum ethmoidale limits the anterior border of the fenestra frontoparietalis. This fenestra is bordered laterally by a thin taenia tecti marginalis and posteriorly by the tectum synoticum (Fig. 5A). At Stage 31, a taenia tecti medialis and a taenia tecti transversalis begin to develop.

A fenestra basicranialis is present at Stage 30. The planum intertrabeculare is better developed in *Hyla geographica* and two pairs of foramina, the foramen caroticum primarium and the foramen craniopalatinum are visible (Fig. 5B). In *H. semilineata*, the fenestra basicranialis is large and only the foramen caroticum primarium is identifiable.

The lateral walls of the braincase are formed by the cartilages orbitales where a large foramen opticum and a smaller foramen prooticum are found. Between the two foramina, a small foramen oculomotorium is present (Fig. 5C).

The capsulae auditivae are spherical, large, about one quarter of the chondrocranial length, and fuse to the posterior floor of the braincase (Fig. 5B, C). A larval crista parotica projects

from the capsulae auditivae laterally (Fig. 5A, C). A processus anterolateralis is present, extending from the anterolateral end of the capsulae auditivae and curving ventrally toward the posterolateral curvature of the palatoquadrate, without fusing with it (Fig. 5A, C). It disappears prior to metamorphosis. A large fenestra ovalis is present, ventral to the processus anterolateralis (Fig. 5C). At Stage 39, the operculum can be seen as a small rounded cartilage inside the fenestra ovalis.

The palatoquadrate is connected to the cranial floor anteriorly by the commissura quadrato-cranialis anterior (Fig. 5B). The larval processus antorbitalis projects dorsolaterally at the level where the commissura quadrato-cranialis anterior attaches to the orbital cartilage (Fig. 5A). In lateral view, this process is hidden by the high, triangular-shaped, round-tipped processus muscularis quadrati that extends dorsally (Fig. 5C). Anterior to the commissura quadrato-cranialis anterior lies a robust pars articularis quadrati, which articulates broadly with the posterior margin of Meckel's cartilage (Fig. 5A). A triangular processus quadrato ethmoidalis extends medially from the pars articularis quadrati (Fig. 5A, B). Posteriorly, the palatoquadrate attaches to the cranial floor by the processus ascendens, which has a low attachment to the orbital cartilage at the level of the pila antotica (Fig. 5B, C). The commissura quadrato-cranialis anterior and the processus ascendens have eroded by Stage 43 in *Hyla geographica* and Stage 44 in *H. semilineata*. Ventral to the processus muscularis quadrati there is a concavity in the palatoquadrate, the fascies articularis quadrati, which is the articular surface for the ceratohyal.

The visceral skeleton consists of paired ceratohyales, the planum hyobranchiale, and four ceratobranchialia (Fig. 5D). The ceratohyales are thick and wide plates of cartilage that underlie the floor of the buccal cavity and are connected ventromedially by the pars reuniens. The ceratohyales have a large processus anterior and a small processus anterolateralis hyalis. The copula I is a slender and elliptic bar of cartilage located anteriorly to the pars reuniens. The copula II extends from the pars reuniens and articulates with the plana hyobranchiales laterally. Ventrally, it has a large cylindrical processus urobranchialis. The plana hyobranchiales are continuous with the first and fourth ceratobranchialia. The ceratobranchialia are connected posteriorly by the commissurae terminales. A slender processus branchialis between the second and third ceratobranchialia is present but not closed. The first three ceratobranchialia each bear a spiculum that projects dorsally at their proximal ends.

DISCUSSION

The internal oral anatomy of the *Hyla geographica* and *Hyla semilineata* tadpoles is overall characteristic of type IV suspension feeder tadpoles (Wassersug, 1976, 1980). Some features, like the number of prepocket papillae and the number of lateral roof papillae have little systematic value because they are highly variable intraspecifically or may be distorted during preparations (Wassersug and Heyer, 1988). However, the number, size, shape, and position of the lingual papillae, located on the buccal floor, are among the characteristics that exhibit low intraspecific variation and may therefore have a taxonomic value (Wassersug and Heyer, 1988). The larvae of these two species can be distinguished by their lingual papillae: the distance between the lingual papillae was three times greater in *H. geographica*, and their relative size was less than half that of *H. semilineata*. Furthermore, these characteristics were constant among the individuals examined for each species.

The detailed study of the internal oral anatomy of these two species revealed results that help to distinguish between them, which is not possible by simply examining their oral discs. Our observation of the oral disc of *Hyla geographica* and *H. semilineata* agrees with earlier reports (Bokermann, 1963; Kenny, 1969; Duellman and Lescure, 1973). The dental formulas previously described were observed in the specimens of both species examined in this study, but cannot be used to distinguish the two taxa since they changed throughout development and were intraspecifically variable. The formula 2(2)/4 described by Bokermann (1963) and Kenny (1969) was observed in some individuals examined by us, although it was not constant throughout development. The rows of denticles that we found beneath the lateral folds of papillae, in some individuals of *H. geographica* and *H. semilineata* between Stages 27–30, could be interpreted as an interrupted third upper row, in which case, the dental formula could then be described as 3(1)/5 as observed by Duellman and Lescure (1973).

Larvae of *Hyla geographica* and *H. semilineata* have indistinguishable chondrocrania. The chondrocranium of only three other species of *Hyla* have been described with enough detail for comparison to these species, namely, *Hyla pulchella andina* (Lavilla and Fabrezi, 1987), *H. lanciformis* (de Sá, 1988) and *H. nana* (Fabrezi and Lavilla, 1992). Although there are slight differences between these three species and those studied in this paper, in general the chondrocrania are similar and are probably typical for the genus.

The tadpole cranium of *Hyla geographica* and *H. semilineata* has a few differences in its developmental sequence. At Stage 30, the foramen craniopalatinum is identifiable in the floor of the braincase in *H. geographica*, but not in *H. semilineata*. Apart from differences in the lingual papillae between the two taxa, the other noticeable difference between *Hyla geographica* and *H. semilineata* is that the latter is about 50% larger at most developmental Stages. The principal component analysis revealed that size is the most distinguishable variable and accounts for most of the variation between the species and that the second principal component, a shape factor, showed hardly any variation. When the data were corrected for size, the principal component analysis also showed less variation between the species. However, this PCA revealed that, although *H. semilineata* is larger than *H. geographica* overall, some of its features are relatively smaller. The morphometric differences are in fact statistically significant as could be seen by the MANCOVA performed using size and stage as covariates.

The morphometric differences and the differences in qualitative characters of the buccal floor provide further support for the recognition of two separate taxa based on adult morphology (Silveira and Caramaschi, 1989). Although Duellman (1973) considered *H. semilineata* to be synonymous to *H. geographica*, his distance phenogram showed that populations of *H. geographica* from the State of Amazonas and the State of Pará (North of Brazil) are more closely related to those of *H. geographica* from Ecuador and Trinidad than to the populations from the Atlantic rainforest (Espírito Santo and Santa Catarina States). Even though the precise distribution of *Hyla semilineata* has not yet been established (Silveira and Caramaschi, 1989), the populations in the Atlantic rainforest most likely correspond to *H. semilineata*.

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